

## What is Clarmed is

A polypeptide free of botulinum toxin activity and free of toxoid which 1. induces protective immunity to a type F botulinum toxin

2.

- A polypeptide characterized in that it:b is free of toxoid and
  (a) is free of botulinum toxin activity,
- is free of toxoid, and
- is capable of eliciting, in a mammal, an immunological response that (c) is protective against type F botulinum/toxin.
- A polypeptide according to Claim 1 2 comprising a fragment or a 3. derivative of a heavy chain of a type F botulinum neurotoxin.
- A polypeptide according to Claim 3 wherein said fragment or said derivative 4. is up to 600 amino acids long.
- Claim 3 A polypeptide according to <del>Claims 3 or 4</del> wherein said fragment is selected from the group consisting of:
  - amino acids 848-1/278 of a type F botulinum toxin, (a)
  - amino acids 848/991 of a type F botulinum toxin, (b)
  - amino acids 992-1135 of a type F botulinum toxin, and (c)
  - amino acids 1/136-1278 of a type F botulinum toxin. (d)
- Claim 3 A polypeptide according to Claims 3 or 4 wherein said derivative comprises 6. a dimer of the fragment according to any of (a)-(d) of Claim 5.
- A polypeptide composition for use in manufacture of a vaccine, said 7. composition comprising:
  - a/polypeptide free of toxin activity and capable of inducing, in a (1) mammal, protective immunity against a botulinum toxin; and

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(2) a polypeptide adapted to facilitate or enhance purification of the composition.

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A polypeptide composition according to Claim 7 wherein the composition comprises a fusion protein of (1) and (2).

A polypeptide composition according to Claim 7 or 8 comprising:-

- (1) a polypeptide according to any of Chaims 1-6; and
- (2) a polypeptide adapted to bind to a chromatography column.
- 10. A polypeptide composition according to any of Claims 7-9 comprising a polypeptide adapted to bind to an affinity chromatography column.
- 11. A polypeptide according to Claim 8 comprising a fusion protein of:-

- (a) amino acids 848 to 1278 of a type F botulinum neurotoxin, with
- (b) a purification moiety.

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A vaccine comprising a pharmaceutically acceptable carrier and a polypeptide according to any of Claims 1.6 or a polypeptide composition according to any of Claims 7-11.

- 13. A recombinant DNA encoding a polypeptide according to any of Claims 1-6 or a polypeptide composition according to any of Claims 7-11.
- 14. A method of producing a polypeptide according to any of Claims 1-6-or a polypeptide composition according to any of Claims 7-11 comprising the steps of:

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- (a) expressing in a host cell a DNA encoding a fusion protein, said protein being a fusion of (i) a fragment or derivative of a type F botulinum toxin, and (ii) a moiety adapted to bind to a chromatography column,
- (b) obtaining from said host cell an extract comprising the fusion protein, and
- (c) purifying the fusion protein using a chromatography column.
- 15. A method according to Claim 14 wherein the chromatography column is an affinity chromatography column and the fusion protein is removed from the column by elution with a substrate.
- 16. A method according to Claim 14 er 15 further comprising cleaving the fusion protein and retaining the toxin fragment or derivative.
- 17. A method of making a pharmaceutical composition comprising:
  - (a) expressing in a host cell a DNA encoding a fusion protein, said protein being a fusion of (i) a polypeptide free of toxin activity and capable of inducing protective immunity against a botulinum toxin, and (ii) a purification moiety adapted to bind to a chromatography column,
  - (b) obtaining from said host cell an extract comprising the fusion protein,
  - (c) purifying the fusion protein using chromatography column,  $An\phi$
  - (d) incorporating the purified fusion protein into a pharmaceutical composition.
- 18. A method according to Claim 17 wherein said purification moiety binds to an affinity chromatography column.
- 19. A pharmaceutical composition comprising:
  - (a) a fusion protein, said protein being a fusion of (i) a polypeptide fee of toxin activity and capable of inducing protective immunity against a botulinum toxin, and (ii) a polypeptide adapted to bind to a chromatography column; and

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A pharmaceutical composition according to Claim 19 wherein said fusion protein comprises a polypeptide according to any of Claims 1-8.

24

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A pharmaceutical composition according to Claim 19 or 20 wherein the fusion protein comprises a polypeptide adapted to bind to an affinity chromatography column.

- 22. A method of vaccinating a mammal against a botulinum toxin, comprising administering to said mammal avaccine according to Claim 12.
- 23. A method of vaccinating a mammal against a botulinum toxin, comprising administering to said mammal a pharmaceutical composition according to Claims 19-21.

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